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Publication date:
2011

Document Version
Publisher's PDF, also known as Version of record

DTU Vet
National Veterinary Institute

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Surveillance of *Campylobacter* ssp. in broiler flocks by PCR on boot sock samples

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Background: Surveillance of *Campylobacter* in broilers is traditionally based on microbiological culture of faecal samples from cloacae or caecum collected at the abattoir. Due to long time needed for analysis, no intervention can be taken in order to prevent meat from positive flocks for being sold on the fresh market. Furthermore, many efforts is taken to increase biosecurity at farm level and the number of positive flocks are decreasing, however the negative flocks may be cross-contaminated during slaughter. To know the Campylobacter status of the broiler flock before arriving at the abattoir, a surveillance system based on *in-flock* sampling using one pair of boot sock combined with PCR detection was established and validated.

❖Methods

❖One pair of sock samples were collected by the farmer 2-10 days prior to slaughter and 10 cloacal swabs were collected at the abattoir.

❖All samples were submitted to the laboratory using ordinary mail.

❖Fecal material was eluted from the socks or the swabs, and following the addition of a internal purification and PCR control (*Yersinia ruckerii*), total DNA was purified using magnetic beads.

❖DNA was added to a multiplex mastermix containing genus specific primer pairs targeting 16S rDNA in *Campylobacter* ssp. and *Yersinia ruckerii.*

❖PCR products were visualized by gel electrophoresis

Using 15 cm of tube gauze mounted on each boot, fecal material was collected by the farmer during inspection in the broiler flock. At the laboratory fecal material was released from the sock by adding water, and following sedimentation of large organic material, one ml of the supernatant was used for DNA purification.

❖Results

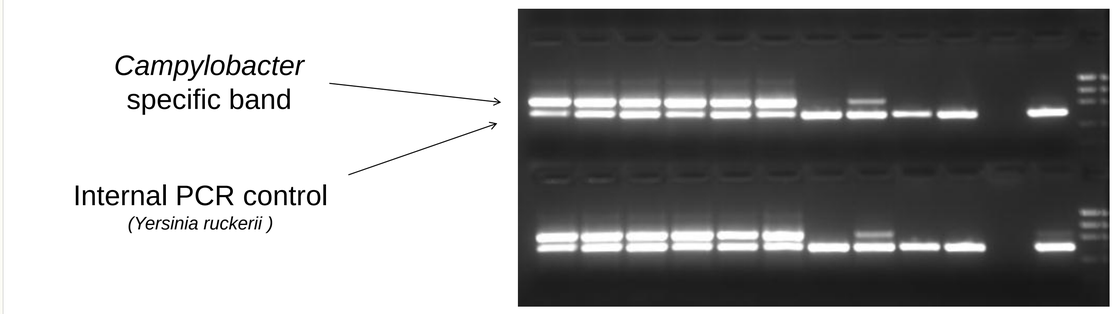
➢A genus specific multiplex PCR assay to detect *Campylobacter* ssp. in fecal samples was established and validated.

➢The addition of an aquatic bacteria, *Yersinia ruckerii,* to the sample served as internal control both during DNA purification and through out the PCR detection.

➢Sensitivity of the assay was 100-300 CFU/ml when negative samples were spiked with *Campylobacter jejuni* before DNA purification, corresponding to 10^3 CFU / gram feces.

➢When sampling was preformed 6-10 days prior to slaughter, we found a correlation between the socks and the swab at slaughter below 67%.

➢For sampling on day 5 or less we found a correlation of 80% or higher.



Gel picture showing 10 sock samples tested in duplicate for *Campylobacter* ssp. In negative samples, only the internal PCR control is amplified.

❖Conclusions

➢With the combination of a sensitive PCR method and boot sock sampling, it is possible to detect *Campylobacter* in the broiler flock.

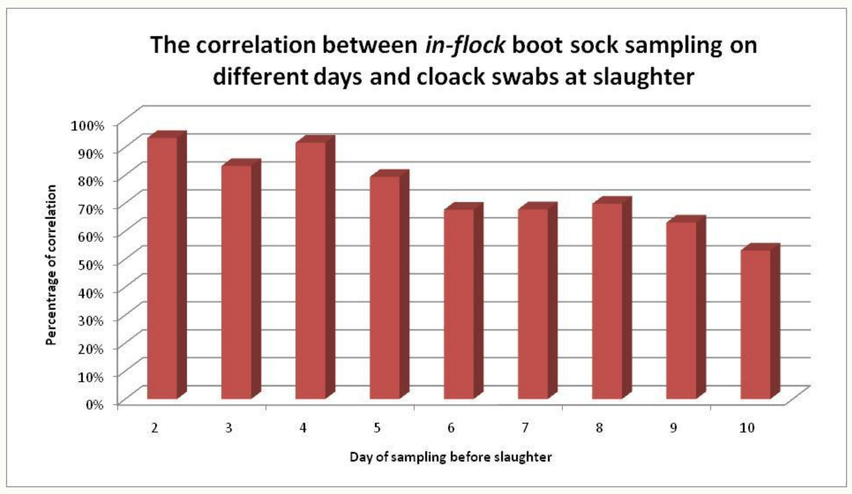
➢Boot socks is an easy and low cost sampling method. It can be preformed by the farmer, and socks may be submitted by ordinary mail. At the laboratory the analysis can be performed within the same day.

➢Due to colonisation dynamics and late introduction of *Campylobacter* in the flock, the predictive value of sock sampling increases when approaching the day of slaughter.

➢The lower predictability of the test early in the growing period indicates that if testing is preformed before day 32, a higher number of socks needs to be taken in each flock.

❖Impact

➢By knowing the status of the flock before it reaches the abattoir, negative flocks may be slaughtered before positive, and the number of *Campylobacter* in meat from positive flocks may be reduced by freezing or marinating.



The closer the sock sample is taken to the day of slaughter, the higher is the correlation with the slaughter house sample.

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